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# Lichen colonization and associated deterioration processes in Pasargadae, UNESCO world heritage site, Iran



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# ABSTRACT

Knowledge on lichen and microbial colonization as well as associated biodeterioration processes of the stone cultural heritage is needed to establish proper conservation programs, but is still poor for stonework in semi-arid regions. In this study, lichen diversity was characterized on seven monumental buildings of the Pasargadae UNESCO-world heritage site (Iran). The risk of biodeterioration processes associated to lichen occurrence on two types of limestones, and the lichen resilience to mechanical cleaning intervention were examined. Physico-chemical substrate features and climatic conditions, combined with the agricultural surrounding and tourist disturbance, supported a pervasive colonization by species-poor epi- and endolithic communities, and fast recolonization processes by nitrophytic species after mechanical removal. The endolithic growth of some lichens and the penetration of hyphal structures of epilithic ones, examined by light and electron microscopy, were associated to stone disintegration and dissolution at the lichen-rock interface. Endolithic cyanobacteria were detected under lichen thalli, likely contributing to deterioration processes. Colonization and deterioration patterns did not appear peculiar with respect to previous investigations on similar communities in different climatic regions, and were mostly related to the different examined lithologies, indicating lichens as harmful biodeteriogens of the sedimentary rock materials used in the stone cultural heritage of semi-arid regions. © 2017 Elsevier Ltd. All rights reserved.

#### 1. Introduction

The archaeological sites and cultural heritage properties, besides being a representation of different civilisation period, positively involve the tourism industry and economy growth of a country (Csapó, 2012). The importance of their conservation and restoration represents an issue that has nowadays assumed a world-wide interest (Harrison and Hitchcock, 2005; Timothy and Nyaupane, 2009). Properly, the colonization by biodeteriogenic organisms is enumerated within the most acute threats for the preservation and conservation of stonework (Caneva et al., 2008; Scheerer et al., 2009). Recently, an increase of their impact has been hypothesised as potentially related to the ongoing global changes in climate and environmental/pollution conditions (Berenfeld, 2008; Viles and Cutler, 2012). Such changes may indeed determine dramatic shifts in stone bioreceptivity and colonization dynamics, and a modification of the biotic interactions with the lithic substrates in areas including vulnerable heritage structures such as the Mediterranean region, Middle East, Caribbean and Southern Africa (Gómez-Bolea et al., 2012; Viles and Cutler, 2012).

A remarkable role of lichens as biological weathering agents of rock substrata has been long recognized and widely documented on both natural outcrops and the outdoor stone cultural heritage (Adamo and Violante, 2000; Ascaso et al., 2002; De los Ríos et al.,

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2009; Gazzano et al., 2009; Seaward, 2015). Epilithic and endolithic lichens can indeed mechanically and chemically damage both the rock surface and interior, mostly because of the adhesion, penetration and changes of volume due to hydratation-dehydratation processes of hyphal structures and the release of metabolites having acidic and/or chelating functions (Ascaso et al., 2002; De los Ríos and Ascaso, 2005). Although lichens represent the most prominent component of lithobiontic communities, biodeterioration processes in a given site are rarely caused only by them. Associated to lichens, but also forming independent communities, different microorganisms such as cyanobacteria, green algae, freeliving fungi and heterotrophic bacteria, contribute to the biodeterioration process, similarly growing both epilithically and endolithically (Bjelland et al., 2011; De los Ríos et al., 2012).

Some investigations showed that in certain conditions lichens may exert bioprotection on the stone surfaces (Carter and Viles, 2005). Bioprotective effects were documented on both natural and man-made mineral substrates under aggressive environmental conditions (Favero-Longo et al., 2009a; Mc Ilroy de la Rosa et al., 2013), for example where high atmospheric pollution occurs or in coastal areas with risk of salt-induced weathering (Mikuláš, 1999; Carballal et al., 2001). However, generalized bioprotective actions of lichens are still retained a controversial subject, needing additional studies (Mc Ilroy de la Rosa et al., 2013).

Due to their efficiency in accumulating nutrients and their resistance to desiccation and temperature extremes and fluctuations, lichens colonize rock substrates in a wide range of habitats, including those normally hostile to other life forms (e.g. De los Ríos et al., 2014). Accordingly, investigations on lichen interactions with the lithic substrate have been performed through a wide geographical range, from polar to tropical areas, revealing different bioweathering and bioprotection patterns depending on different climate conditions, lithologies and lichen communities (Seaward, 2015). In parallel, lichen colonization of stonework has been also sparsely explored up to remote areas (St. Clair and Seaward, 2004), but many combinations of "lithology × lichen community × climate conditions" have hitherto been understudied, even where lichen occurrence has been generally recognized as a potential threat in conservation/restoration plans.

Lichen colonization of the stone cultural heritage in arid/semiarid environments has been less deeply characterized respective to that in the temperate and Mediterranean bioclimatic areas (Mohammadi and Krumbein, 2008). The poorness of information appears particularly serious as the few available reports revealed extremely contrasting effects of the lichen colonization, varying from severe deterioration (Ascaso et al., 2004; Knight et al., 2004; Paradise, 2013) to significant bioprotection (Souza-Egipsy et al., 2004; Özvan et al., 2015). Counteracting processes below the same lichen species were even reported in the case of the endolithic *Verrucaria rubrocincta* on limestone, actively deteriorating its substrate, but concomitantly forming a micrite layer which mitigates the destructive effects (Garvie et al., 2008).

In Iran, five world-heritage sites [Armenian Monastic Ensembles of Iran (West and East Azerbaijan provinces), Bisotun (Kermanshah province), Pasargadae (Fars province), Persepolis (Fars province), Takht-e Soleyman (West Azerbaijan province)] built by stone materials are heavily colonized by lichens (Sohrabi, personal observation). However, although the diversity of lichens and lichenicolous fungi has been recently surveyed in the natural environments of different areas of Iran (e.g. Seaward et al., 2004; Westberg and Sohrabi, 2012), the characterization of the lichen colonization of monuments is largely deficient. Mohammadi and Krumbein (2008) remarkably explored biodeterioration of Persepolis monuments, where they generally described hyphal penetration patterns and pitting effects of the epi- and endolithic lichen components. However, they did not associate the observed phenomena with the different lithologies characterizing the archaeological site and they did not analyze the lichen diversity to properly correlate different species with the observed penetration/deterioration patterns. On the other hand, in the recent years, the lichen diversity has been surveyed on the brick surfaces of the Gonbad-e Qabus, UNESCO world Heritage site (Sohrabi et al., unpublished technical report) and in other minor sites (e.g. on the Tangivar stone inscriptions: Sohrabi and Abbas Rouhollahi, 2012), but related deterioration patterns were not investigated. Lichen colonization has been also recently listed as a risk factor for the conservation of the World Heritage Site of Pasargadae, which is located in a rural plain of South-Western Iran and notably includes the Mausoleum of Cyrus the Great (Rafiee-Fanood and Mehdizadeh-Saradi, 2013). On this latter monument, conservation and restoration interventions were performed in 2006, including the cleaning of the rock surfaces by mechanical methods (water and plastic or wire brushes). However, lichen diversity and the associated deterioration patterns had been not preventively examined to evaluate their effective impact on the rock surfaces.

In this paper, we first survey (a) the lichen diversity on the stone monuments of the World Heritage Site of Pasargadae and we examine (b) the lichen-lithic substrate interface of epilithic and endolithic lichen species to infer their potential effect on limestones with different quartz contents. The colonization patterns and associated mineral-microorganism interactions observed in Pasargadae are then discussed in comparison to those previously reported from arid/semi-arid regions to the most studied Mediterranean/temperate regions, where lichens have been variously associated to biodeterioration or bioprotection effects.

### 2. Material and methods

#### 2.1. Study site

The archaeological site of Pasargadae is located in the Madar-e-Soleyman rural district, Pasargadae county, in the valley of the river Polvar, on the Dasht-e Morghab (the plain of Morghab), approximately 25 km long and 12 km wide (N30° 11' 37.788" E53° 10' 2.244) (Fig. 1). The site, one of the oldest residences of the Achaemenid kings, founded by Cyrus the Great (r. 559-530 BCE), was inscribed in 2004 in the UNESCO World Heritage List (Mozaffari, 2014; http://whc.unesco.org/en/list/1106). Its 2  $\times$  3 km<sup>2</sup> area includes seven monumental buildings, (Fig. 1C, 1-7 locations). A beige limestone (<5% quartz content, BL) from the Sivand Quarry (c. 30 km far from the site), also used in Persepolis, was mostly used in the Cyrus Mausoleum and in the other buildings, but a more darkgrey arenaceous variety of limestone (15–20% quartz content, GL), and other lithologies were also abundantly used (Bonazza et al., 2007; Emami, 2010; Mozaffari, 2014). In BL, calcite clasts are embedded in a very fine calcite matrix (diam. c. 1 µm), while in GL, calcite and quartz grains (diam. c. 50-100 µm) are embedded in a slightly coarser calcite matrix (diam. c.  $1-5 \mu$ m), determining a higher porosity (Supplementary material 2).

Mean annual rainfall for Pasargadae is over 350 mm, the prevailing direction of rain-bearing winds being west to southwest (Talebian, 2014).

#### 2.2. Lichen diversity survey

Lichen diversity was surveyed in the period 2005–2015 on the following monumental buildings in Pasargadae (Fig. 2): (A) The Mausoleum of Cyrus the Great, (B) The Madrasseh or Mozafari Caravanserai, (C) Gate R (Gate House), (D) Palace P (Residential Palace), (E) Palace S (Audience Hall), (F) The Zendan (Zendan-e

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